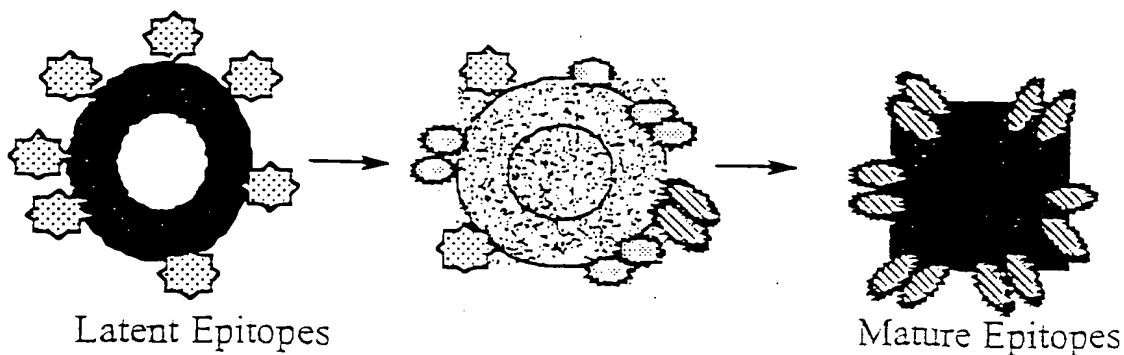
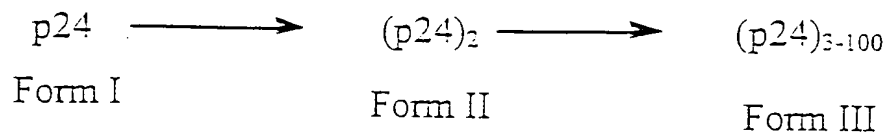


Scheme 1. Conformational search and chemical conversion during rHBsAg maturation

A. Schematic Representation for Epitope Evolution

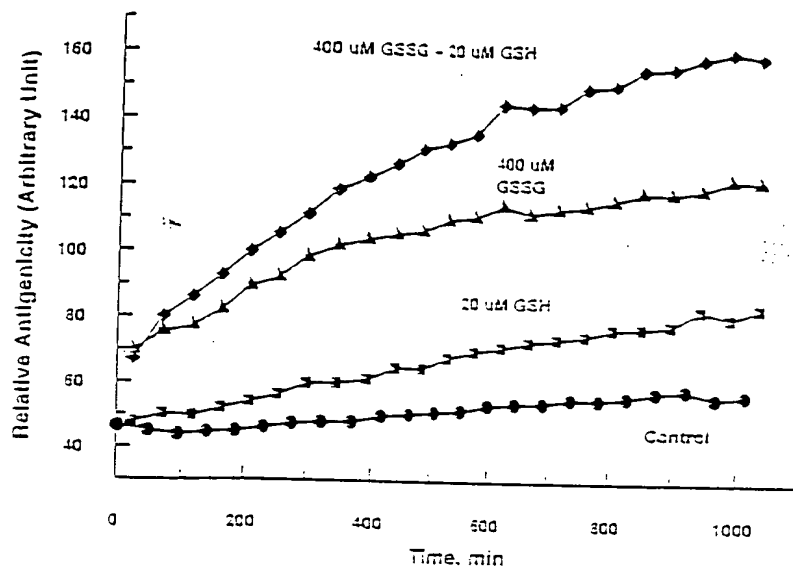


B. Chemical Conversion - Interchain Disulfide Bond Cross-linking

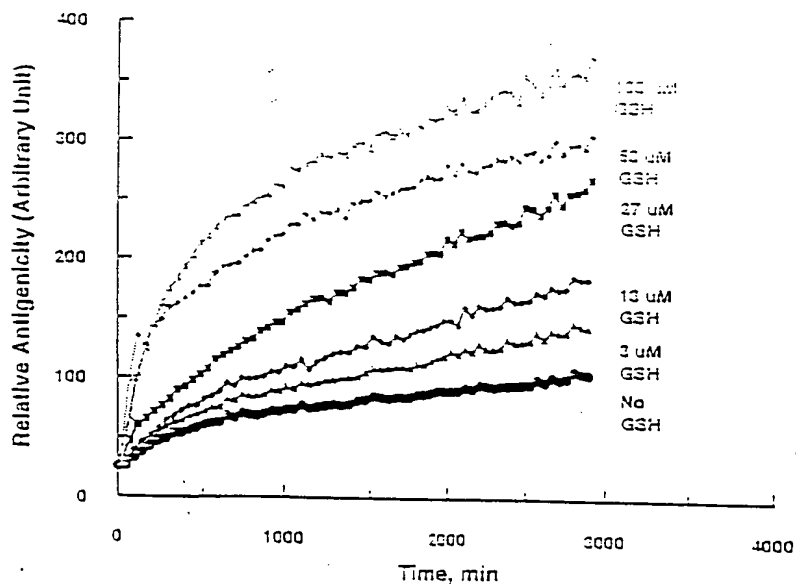


## FIG 2

Glutathione-mediated maturation of rHBsAg at 37 °C. Top: Synergistic effects of GSH and GSSG; Bottom: Better conformation of HBsAg can be achieved by higher concentration of GSH.



GSH-Catalyzed Unscrambling of Disulfide Bond(s) During HBsAg



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	I		IO		20				30				40				50				60																																								
A1.2	D	I	V	L	T	Q	S	P	A	I	H	S	A	S	F	O	E	K	V	T	M	T	C	S	A	S	S	S	V	S	Y	M	Y	*	H	Y	Q	Q	K	P	O	S	S	P	R	L	L	I	F	I	O	T	S	N	L	F	S	G	V	P	V

[illegible]

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[illegible][illegible]

## FIG 4

## Flow Chart

In Vitro Relative Potency Assay

DAY 1

Pre-treat standard and samples - Section III.A.

Dispense 200 mcL of each treated standard or sample dilution into wells↓  
Dispense 200 mcL of the Diluent Control and the Kit Controls into replicate wells↓  
Add 50 mcL of conjugate to wells↓  
Tap tray gently to mix↓  
Add one bead to each test well↓  
Incubate trays for 12-20 hours at 20-28°C

DAY 2

↓  
Wash beads 1 cycle with  $14 \pm 3$  mL distilled water↓  
Transfer beads to tubes↓  
Add 300 mcL of OPD substrate solution↓  
Incubate 30 to 35 minutes at 20-28°C in the dark↓  
Add 1 mL of 1N Sulfuric acid to each tube↓  
Read tubes in Quantumatic™ within 2 hours

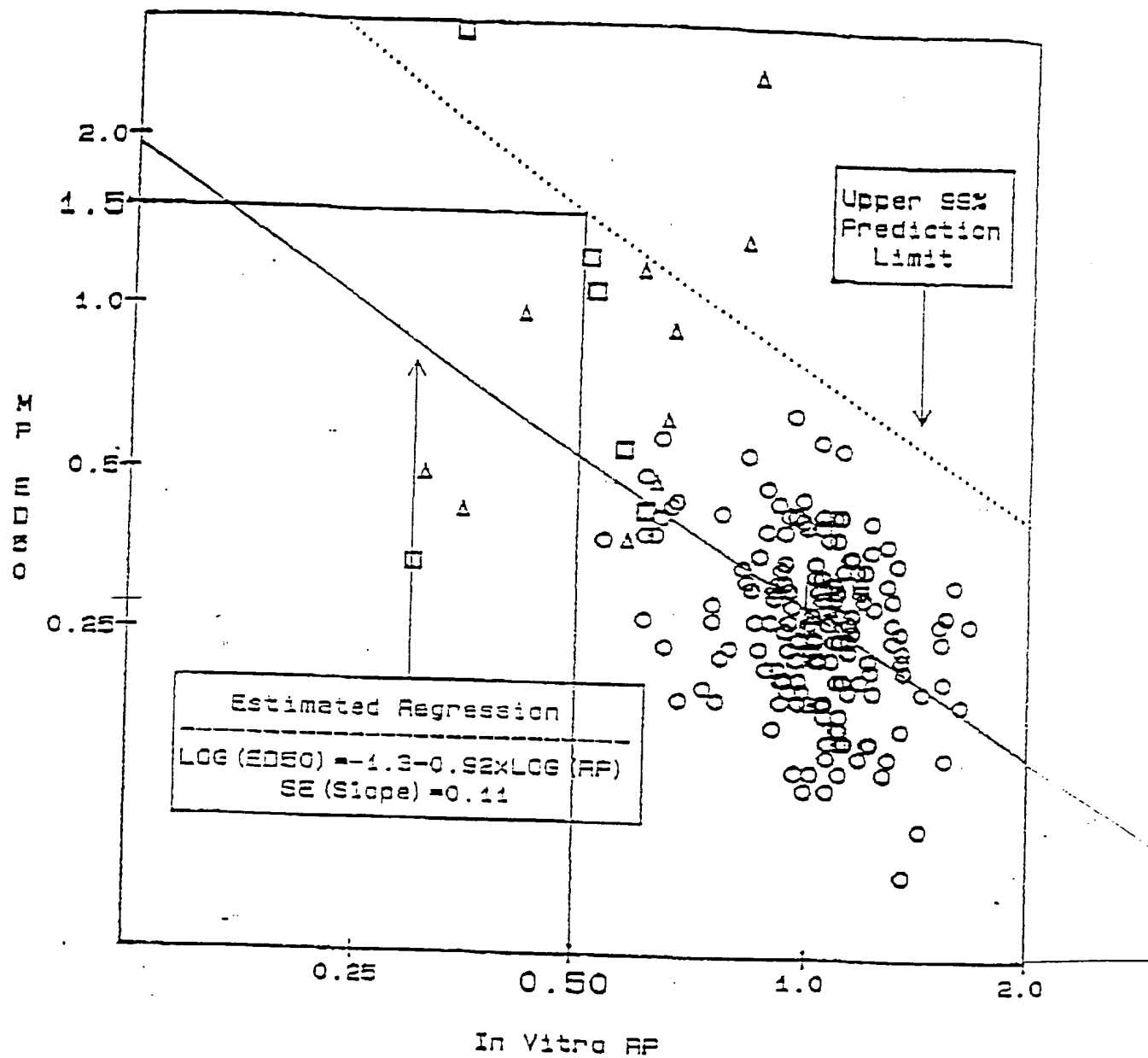
- Read Kit Controls blanked on substrate.

- Read Standards and test blanked on sample diluent.

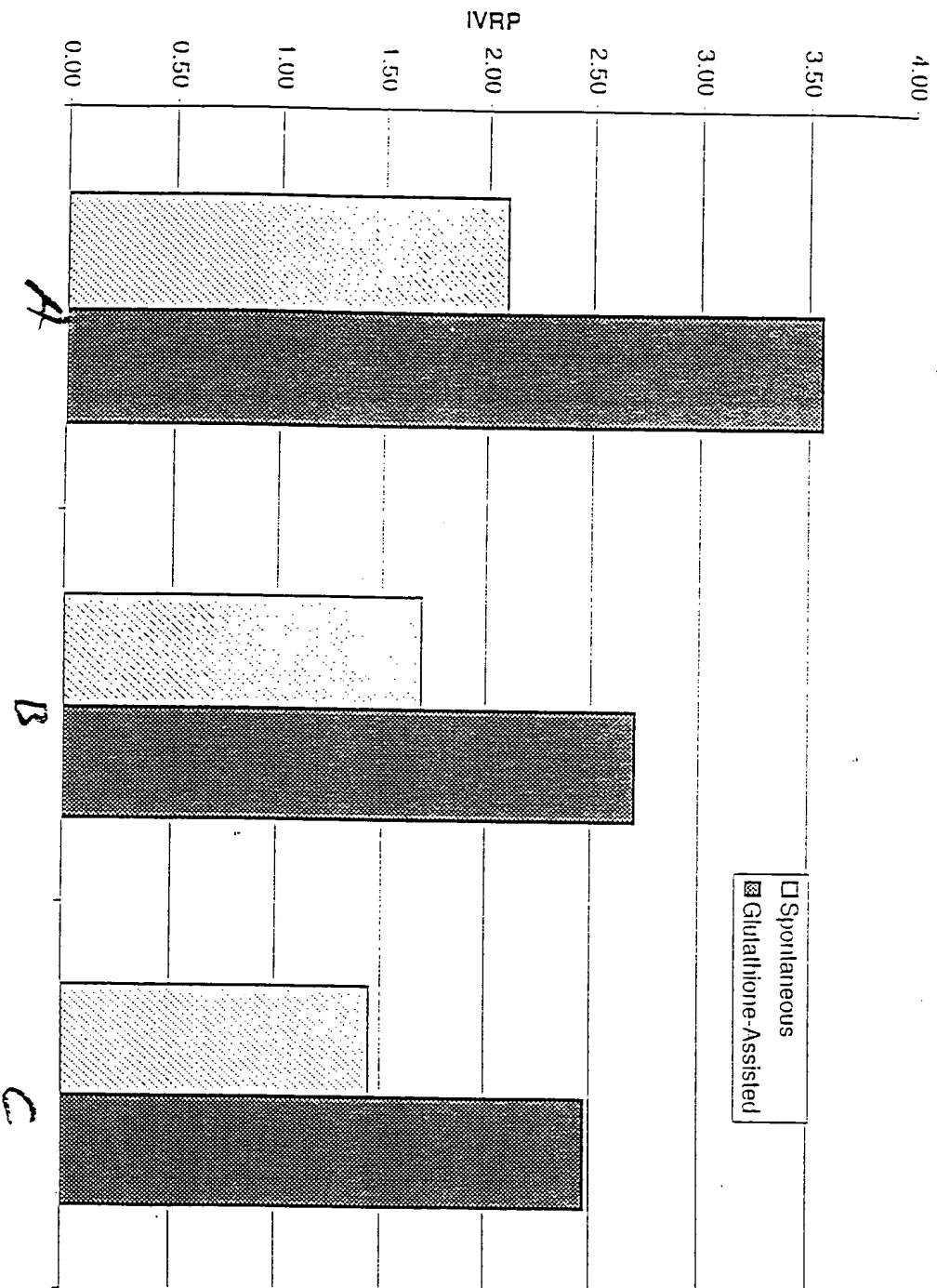
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FIG 5

In Vitro Relative Potency Specification  
Corresponding to Mouse Potency Specification

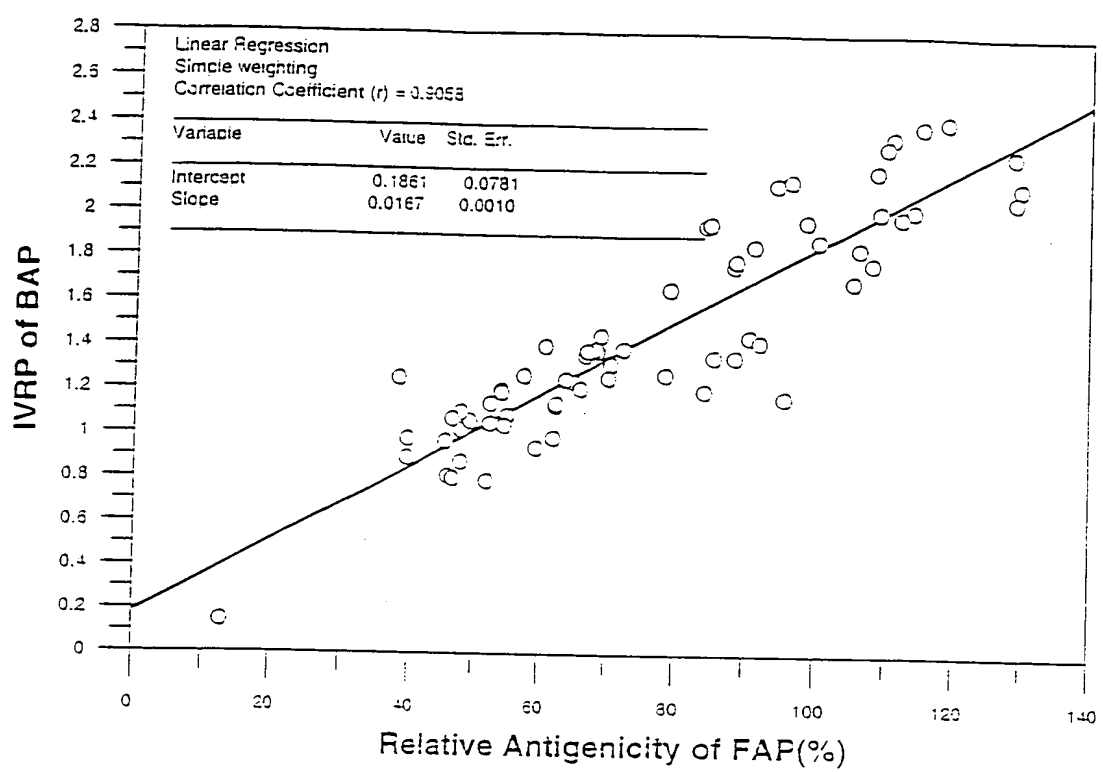


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FIG 7



Linear relationship between IVRP of BAP vs. relative antigenicity of FAP (%) by EIAcore.